TRAINING UPDATE

Lab Location:	SGMC	**Date Distributed:**	7/1/2019
Department:	Core Lab	**Due Date:**	7/31/2019
		Implementation:	**7/15/2019**

DESCRIPTION OF PROCEDURE REVISION

Name of procedure:

AVOXimeter 4000 Whole Blood CO-OXimeter SGAH.C131 v3

Description of change(s):

Section	Reason
3.1, 3.2	Add syringe is preferred, and microtainer is **not** an acceptable collection container

This revised SOP will be implemented on July 15, 2019

Document your compliance with this training update by taking the quiz in the MTS system.

Technical SOP

Title	**AVOXimeter 4000 Whole Blood CO-OXimeter**
Prepared by	Judy Codling / Anne Schoonover Date: 2/11/2013
Owner	Robert SanLuis Date: 2/11/2013

Laboratory Approval	**Local Effective Date:**	
Print Name and Title	**Signature**	**Date**
Refer to the electronic signature page for approval and approval dates.		

Review		
Print Name	**Signature**	**Date**

TABLE OF CONTENTS

1. TEST INFORMATION

Assay	Method/Instrument	Local Code
Carboxyhemoglobin Arterial	AVOXimeter 4000	ACOHG
Carboxyhemoglobin Venous		VCOHG
Methemoglobin Arterial		AMEHG
Methemoglobin Venous		VMEHG

Synonyms/Abbreviations
Carboxy Hgb Arterial/Venous, Met Hgb Arterial/Venous, % COHb, % MetHb

Department
Core Lab

2. ANALYTICAL PRINCIPLE

The ITC AVOXimeter 4000 is a whole blood oximeter that performs individual measurements of carboxyhemoglobin and methemoglobin on heparinized or EDTA anti-coagulated whole blood. The sample is manually injected into a disposable cuvette and inserted into the AVOXimeter. The AVOXimeter illuminates the sample with multiple wavelengths, records the optical density of the sample at multiple wavelengths, and computes the results. Reading on multiple wavelengths reduces the interference from dyshemoglobins, fetal hemoglobins, bilirubin, and hemolysis.

3. SPECIMEN REQUIREMENTS

3.1 Patient Preparation

Component	Special Notations
Fasting/Special Diets	N/A
Specimen Collection and/or Timing	Samples should be obtained from an arterial line, an arterial puncture, or venipuncture ~~or capillary~~. Use of a syringe is preferred.
Special Collection Procedures	Care should be taken to prevent the introduction of air into the sample when it is drawn. When using a syringe, expel all air bubbles from the syringe and cap and seal the end of the syringe.

3.2 Specimen Type & Handling

Criteria		
Type	**-Preferred**	Arterial: Lithium or Sodium heparinized whole blood Venous: EDTA or Sodium heparinized whole blood
	-Other Acceptable	Arterial: EDTA whole blood Venous: Lithium heparinized whole blood
Collection Container		Syringe (preferred) or vacutainer tube (light green, dark green or purple top). Microtainers (bullets) are not acceptable.
Volume	**- Optimum**	100 µl
	- Minimum	60 µl
Transport Container and Temperature		Collection container at room temperature
Stability & Storage Requirements	Room Temperature:	Specimen should be tested as soon as possible after collection.
	Refrigerated:	Not acceptable
	Frozen:	Not acceptable

Form revised 2/02/2007

Criteria	
Timing Considerations	A freshly collected sample is optimal
Unacceptable Specimens & Actions to Take	Any specimen other than heparinized (Li or Na) or EDTA anti-coagulated whole blood. Any clotted specimen. Reject the specimen and have the patient redrawn. Cancel the test with the appropriate LIS English text code.
Compromising Physical Characteristics	Evidence of clotting and/or visible hemolysis. Reject the specimen and request a recollection. Cancel the test with the appropriate LIS English text code.
Other Considerations	**Prior to analysis, the sample should be free of any air bubbles and mixed by rolling the syringe between the outstretched palms of both hands for 10 seconds. Invert the syringe and repeat mixing.** Expel a small amount of blood sample into an absorbent surface. Samples collected at WAH are sent STAT to SGMC.

NOTE: Labeling requirements for all reagents, calibrators and controls include: (1) Open date, (2) Substance name, (3) Lot number, (4) Date of preparation, (5) Expiration date, (6) Initials of tech, and (7) Any special storage instructions. Check all for visible signs of degradation.

4. REAGENTS

The package insert for a new lot of kits must be reviewed for any changes before the kit is used. A current Package Insert is included as a Related Document.

4.1 Reagent Summary

Reagents / Kits	Supplier & Catalog Number
AVOXimeter 4000 cuvettes	ITC QV8

4.2 Reagent Preparation and Storage

Reagent	AVOXimeter 4000 cuvettes
Container	Packaged with a desiccant in the plastic bag
Storage	Store in a closed bag at room temperature (15-30°C)
Stability	Within manufacturer's expiration date as long as the desiccator's indicator is blue.
Preparation	Cuvette must be kept dry at all times. Before analyzing samples, take from the re-sealable bag only as many cuvettes as needed for immediate use. Promptly re-seal bag containing cuvettes and desiccant.

5. CALIBRATORS/STANDARDS

Calibration is performed by the manufacturer and is stable for five or more years.

Calibration Verification will be performed by:
1. Analyzing the Optical QC Filters on a daily basis. This verifies the calibration and confirms that the optics are clean.
2. Analyze liquid Calibration Verification Controls semi-annually. This verifies the calibration, and checks the cuvette storage conditions and the function of the cuvettes.

5.1 Calibrators/Standards Used

Calibrator	Supplier and Catalog Number
Calibration Verification Controls	RNA Medical CVC 223

5.2 Calibrator Preparation and Storage

Calibrator	RNA CVC 223
Preparation	Allow to equilibrate at room temperature for 30 minutes before testing
Storage/Stability	5-8°C, until manufactures expiration date

5.3 Calibration Verification Procedure

Criteria	Special Notations
Frequency	Every 6 months
Tolerance Limits	All results must be within the manufactures acceptable range
Procedure	Follow section 8.3
Dilutions	None
-Graph Type	N/A

6. QUALITY CONTROL

6.1 Controls Used

Controls	Supplier and Catalog Number
RNA Co-Oximeter control	RNA Medical QC 527-1 & QC 527-3
Optical QC filters (yellow & orange)	ITC 4-QCYO

6.2 Control Preparation and Storage

Control	RNA Co-Oximeter control
Preparation	Allow to equilibrate at room temperature (20-25ºC) for at least 4 hours before testing. Before use, hold the ampule at the top and bottom (with forefinger and thumb) and shake for 10 seconds to mix.

Storage/Stability	**Unopened**: 2-8°C, until manufactures expiration date. May also be stored at room temperature (up to 25°C) for 12 months provided the labeled expiration date is not exceeded. **Opened**: stable for one (1) hour

Control	Optical QC filters
Preparation	None
Storage/Stability	Room temperature

6.3 Frequency

- Optical Controls are tested and documented with each patient run.
- A minimum of two levels of liquid QC material will be analyzed weekly.
- If desiccant indicator is pink at 20, 30, or 40 liquid QC must be run and within range limits supplied be QC manufacturer prior to releasing patient results.

6.4 Tolerance Limits and Criteria for Acceptable QC

If a result is outside of the range limits supplied be QC manufacturer, this represents an unsuccessful quality control test.

- **DO NOT release patient results when controls do not meet tolerance limits.** Investigate and take corrective action for all unacceptable controls. Corrective action must be documented.
- Check the lot numbers of the control and cuvettes to ensure that the correct lot number was entered. Check the Cuvette Cal Code in the instrument with the one printed on the cuvette package. Check the desiccant indicator in the cuvette package. If the desiccant indicator is pink at 20, 30, or 40 liquid QC must be run and in range prior to releasing patient results.
- Re-run control
- Repeat the test with a new ampule. If the results are within range proceed with patient testing.
- If results are still outside of range limits, contact ITC and discontinue patient testing until the issue is resolved.

6.5 Documentation

Document results on the QC or Patient Testing log, as appropriate.

6.6 Quality Assurance Program

- Each new lot number of reagent or new shipment of the same lot of reagent must be tested with external control materials. Performance of the new lot must be equivalent to the previous lot.

- Training must be successfully completed and documented prior to performing this test. This procedure must be incorporated into the departmental competency assessment program.
- The laboratory participates in CAP proficiency testing. All proficiency testing materials must be treated in the same manner as patient samples.
- Consult the Laboratory QC program for complete details.

7. EQUIPMENT and SUPPLIES

7.1 Assay Platform

AVOXimeter 4000 (including attached temperature probe)

7.2 Equipment

None

7.3 Supplies

- Optical Filters
- Gauze (absorbent surface)
- Syringes (1-3 ml **non-heparinized**) and Plastic Dispense Tips (or Needles)
- Cotton swabs
- Isopropyl Alcohol
- 18 - 20 gauge needles

8. PROCEDURE

NOTE: For all procedures involving specimens, buttoned lab coats, gloves, and face protection are required minimum personal protective equipment. Report all accidents to your supervisor.

8.1	Maintenance
1.	Clean the analyzer's exterior with a disinfecting solution (i.e., isopropyl alcohol, 10% bleach, etc.) after testing.
2.	Document on the QC log.

8.2	Quality Control testing with Optical Filters
1.	Verify the level of control to be tested (Yellow or Orange).
2.	Turn the instrument on. When the "READY" screen appears insert the colored filter into the AVOXimeter 4000.
3.	Indicate sample type as QC, then press *enter*
4.	Indicate QC type as optical, then press *enter*
5.	Indicate whether the filter is orange or yellow, then press *enter*.

8.2	Quality Control testing with Optical Filters
6.	Verify the optical filter color, then press *enter*.
7.	After analysis, transcribe the data into the Quality Control Record File.
8.	Results should be within the range printed on the filter and at the top of the QC form.
9.	Remove the filter. Press *enter*. Instrument will return to the "READY" screen.

8.3	Quality Control testing liquid QC samples
1.	Verify the level of control to be tested (QC Level 1, Level 3 or CVC).
2.	Allow at least 4 hours for QC ampules to equilibrate at room temperature (20-25ºC) prior to testing.
3.	Verify that the Cuvette Cal Code is correct for the cuvettes in use. The calibration code for the cuvette lot is found on the label of the cuvette packaging. The READY screen displays the cal code stored in the analyzer memory. If it is different than the current calcode in use: Press the **Main menu** key**.** Select **Calibration**, **Cuvette Cal Code**, and **Enter New Value.** Enter the correct cal code using the numeric keypad. Press **OK** to enter the code into the analyzer's memory. Press Cancel twice to return to the "READY" screen
4.	Mix the control sample according to the manufacturer's recommendations, approximately ten seconds.
5.	Restore liquid to the bottom of the ampule with gentle tapping. If foam or small bubbles are present, allow ampule to stand until these have come to the surface.
6.	With fingers protected, carefully snap open the ampule.
7.	Contents should be sampled as soon as the ampule is opened. It is necessary to transfer liquid from the ampule to a syringe. Use an 18 - 20 gauge needle on a 1-3 ml **non-heparinized** syringe. Insert the needle to the bottom of the ampule and slowly draw liquid into the syringe.
8.	Remove needle and insert the syringe tip into the cuvette syringe port and inject the QC sample into the cuvette, holding cuvette down at a 45° angle. Inject QC material into the cuvette until the sample reaches the vent patch. Leave syringe attached to cuvette. **Note**: Over injection of QC material will cause the vent patch to bulge outward. If this happens, pull back slightly on the syringe plunger just until the patch flattens.
9.	Check that no air bubbles are present in the sample light path. **Note**: Air bubbles will yield erroneous results. If air bubbles are present in the light pathway, discard cuvette.
10.	Holding the cuvette by the black cap, insert it into the slot of the instrument's front panel.
11.	Insert the cuvette within 30 seconds of filling it. **Note**: A delay of analysis greater than 30 seconds may yield erroneous results.
12.	Enter User ID number using the numeric keypad and press **Enter/On**.

8.3	Quality Control testing liquid QC samples
13.	Indicate sample type as **QC**.
14.	Indicate the level, the QC material lot numbers that were predefined for that level, and verify the Cuvette Lot number or enter the new Cuvette Lot number.
15.	After analysis transcribe the data into the Quality Control Record File.
16.	Results must be within the acceptable ranges.
17.	Remove the cuvette and then press *enter* to return instrument to the "READY" screen.

8.4	Test Run
1.	Verify that the Cuvette Cal Code is correct for the cuvettes in use. The calibration code for the cuvette lot is found on the label of the cuvette packaging. The *Ready* screen displays the cal code stored in the analyzer memory. If it is different than the current calcode in use: Press the **Main menu** key**.** Select **Calibration**, **Cuvette Cal Code**, and **Enter New Value.** Enter the correct cal code using the numeric keypad. Press **OK** to enter the code into the analyzer's memory. Press Cancel twice to return to the "READY" screen
2.	Roll the syringe containing the blood sample between hands periodically inverting the syringe to fully mix the sample. The sample **must be mixed for a full ten (10) second** interval just prior to injection into the cuvette. **Note**: *Poorly mixed samples or those containing clots may cause inaccurate results*.
3.	Expel a small amount of sample from syringe. Insert the syringe tip into the cuvette syringe port and inject the blood sample into the cuvette, holding cuvette down at a 45° angle. Inject blood into the cuvette until the sample reaches the black vent patch. Leave syringe attached to cuvette. **Note**: Over injection of blood will cause the vent patch to bulge outward. If this happens, pull back slightly on the syringe plunger to flatten patch.
4.	Check that no air bubbles are present in the sample light path. **Note**: Air bubbles will yield erroneous results. If air bubbles are present in the light pathway, discard cuvette.
5.	Holding the cuvette by the black cap, insert it into the slot of the instrument's front panel.
6.	Insert the cuvette within 30 seconds of filling. **Note**: A delay in analysis of greater than 30 seconds may yield erroneous results.
7.	Indicate sample type as **Patient.**
8.	Enter the patient identification number using the numeric keypad. Verify the number on the screen and press **Enter/On** to enter the number into the analyzer's memory.
9.	The results will print automatically. Remove the cuvette and then press enter to return the instrument to the "READY" screen.
10.	Record the results on the Patient Testing log and manually enter the results in the LIS.

NOTE: In the event that the test system becomes inoperable, notify supervision or designee for further direction. Patient specimens must be stored in a manner that maintains the integrity of the specimen.

9. CALCULATIONS

None

10. REPORTING RESULTS AND REPEAT CRITERIA

10.1 Interpretation of Data

The AVOXimeter 4000 reports negative values for both carboxyhemoglobin and methemoglobin. All negative values are to be reported as "0".

10.2 Rounding

None required

10.3 Units of Measure

Analyte	Unit
% COHb	%
% MetHb	%

10.4 Clinically Reportable Range (CRR)

Carboxyhemoglobin	0.0 – 75.0%
Methemoglobin	0.0 – 70.0%

10.5 Review Patient Data

Check for unusual patterns, trends, or distributions in patient results (such as an unusually high percentage of abnormal results). Resolve any problems noted before issuing patient reports.

10.6 Repeat Criteria and Resulting

All repeats must replicate the original result within the total allowable error (TEa) of the assay**.**

Values that fall within the CRR may be reported without repeat. Values that fall outside these ranges must be repeated.

IF the result is …	THEN…
≤ the lower level of the CRR	Assure that the blood sample in the cuvette is devoid of bubbles or clots. Repeat the sample. If the result repeats, then report as the lower level of the CRR.
≥ the upper level of the CRR	Assure that the blood sample in the cuvette is devoid of bubbles or clots. Repeat the sample. If the result repeats, do not report without supervisor approval.

LIS resulting:
Function **MEM**
Worksheet: SCH1
Enter Accession number
Enter result
Type (D) to display and review result
Type (A) to accept result

11. EXPECTED VALUES

11.1 Reference Ranges

Carboxyhemoglobin (%COHb)	0.0 – 4.0 %
Methemoglobin (%MetHb)	0.0 – 3.0 %

11.2 Critical Values

Carboxyhemoglobin (%COHb)	>10.0 %
Methemoglobin (%MetHb)	<0.1, > 3.5 %

11.3 Standard Required Messages

None established

12. CLINICAL SIGNIFICANCE

The portable Avoximeter 4000 provides rapid, accurate assessment of the patient's carboxyhemoglobin and methemoglobin status.

Carboxyhemoglobin is a stable complex of carbon monoxide and hemoglobin that forms in red blood cells when carbon monoxide is inhaled or produced in normal metabolism. Large quantities of it hinder delivery of oxygen to the body. Tobacco smoking (through carbon monoxide inhalation) raises the blood levels of COHb by a factor of several times from its normal concentrations.

Normally one to two percent of a person's hemoglobin is methemoglobin; a higher percentage than this can be genetic or caused by exposure to various chemicals and depending on the level can cause health problems known as methemoglobinemia.

13. PROCEDURE NOTES

- **FDA Status**: Approved
- **Validated Test Modifications:** None

1. Never re-use a test cuvette once it has been inserted into the analyzer.
2. Do not remove the syringe from the cuvette until testing is complete and the cuvette has been removed from the analyzer.
3. Discard cuvettes with trapped air bubbles and check for blood clots from patient specimen prior to injection. Prior to analysis the sample should be mixed by rolling the syringe between the outstretched palms of both hands for 10 seconds. Invert the syringe and repeat mixing. Invert the sample syringe and allow air bubbles to rise (tap syringe if necessary). Expel a small amount of blood sample into an absorbent surface until all air has been removed. A sample with clots or air bubbles may yield erroneous results.
4. Common sources of sampling errors:
   - Sample not freshly drawn
   - Introduction of room air
   - Sample was not mixed well enough
   - Air bubbles or clots present in the light path
   - Type of anticoagulant used
   - Overfilling of cuvette
   - Not inserting cuvette into the analyzer in less than 30 seconds

14. LIMITATIONS OF METHOD

14.1 Analytical Measurement Range (AMR)

Carboxyhemoglobin	0.0 – 75.0%
Methemoglobin	0.0 – 70.0%

14.2 Precision

Carboxyhemoglobin	≤ 5.0%
Methemoglobin	≤ 5.0%

14.3 Interfering Substances

An interferent is a substance which if present in significant levels in the blood specimen being analyzed, will produce an error in the result of the analyte being measured.

Interfering substance	%CoHb	%MetHb
Bilirubin	<1%	<1%
Hemolysis	<1%	<1%
Fetal Hemoglobin tHb=13.5 g/dL, Hbf = 100%)	0.06%/%HbF	<1%
Indocyanine green	<1%	<1%

14.4 Clinical Sensitivity/Specificity/Predictive Values

None

15. SAFETY

Refer to your local and corporate safety manuals and Safety Data Sheet (SDS) for detailed information on safety practices and procedures and a complete description of hazards.

16. RELATED DOCUMENTS

1. Laboratory Quality Control Program
2. Laboratory Safety Manual
3. Safety Data Sheets (SDS)
4. AVOXimeter 4000 Quality Control Log (AG.F203)
5. AVOXimeter 4000 Patient Testing Log (AG.F230)

17. REFERENCES

ITC AVOXimeter 4000 Operator's manual

18. REVISION HISTORY

Version	Date	Section	Reason	Reviser	Approval
000	10/28/13	3.2	Change preferred specimen for venous collections to EDTA	L Barrett	R SanLuis
000	10/28/13	16	Add forms	L Barrett	R SanLuis
000	10/28/13	Footer	Version # leading zero's dropped due to new EDCS in use as of 10/7/13.	L Barrett	R SanLuis
1	10/26/17	4,5,6	Remove individual section labeling instructions and add general one	L Barrett	R SanLuis
1	10/26/17	4.2	Update prep instructions	J Negado	R SanLuis
1	10/26/17	6.1	Correct RNA catalog numbers	J Negado	R SanLuis
1	10/26/17	6.2	Update storage and prep for RNA QC	J Negado	R SanLuis
1	10/26/17	8.3	Add step for QC to equilibrate	J Negado	R SanLuis
1	10/26/17	10.5	Move patient review from section 6	L Barrett	R SanLuis
1	10/26/17	10.6	Add LIS resulting instructions	J Negado	R SanLuis
1	10/26/17	13	Update to new standard wording	L Barrett	R SanLuis

Version	Date	Section	Reason	Reviser	Approval
2	6/10/19	3.1, 3.2	Add syringe is preferred, and microtainer is not acceptable	L Barrett	R SanLuis

19. ADDENDA

None